

AMENDMENTS TO THE CLAIMS

This listing of claims will replace all prior versions, and listings, of claims in the application:

Claim 1. (Original) A process for production of plasmid DNA comprising:
(a) selecting a highly productive clonal subtype of a strain of *E. coli* harboring a DNA plasmid, wherein a highly productive clonal subtype exhibits a higher plasmid copy number per cell in comparison to non-selected, transformed *E. coli* clonal subtypes of the same strain; and,
(b) cultivating said highly productive clonal subtype with fed-batch fermentation in chemically-defined medium.

Claim 2. (Original) A process of claim 1, wherein said selection in step (a) comprises:
(a) a first selection step wherein potential highly productive clonal subtypes of a strain of *E. coli* harboring a DNA plasmid are isolated; and,
(b) a second selection step wherein said potential highly productive clonal subtypes isolated in step (a) are tested to determine which of said clonal subtypes are highly productive.

Claim 3. (Original) A process of claim 2, wherein said strain of *E. coli* is DH5.

Claim 4. (Original) A process of claim 2, wherein colonies of said potential highly productive clonal subtypes of step (a) are phenotypically gray on blood agar.

Claim 5. (Currently amended) A process of claim 4, wherein said gray colonies are visible after incubating on blood agar ~~is incubated~~ for about 48 hours at about 30°C.

Claim 6. (Original) A process of claim 2, wherein colonies of said potential highly productive clonal subtypes of step (a) are phenotypically cream on chemically-defined agar medium after incubating said *E. coli* until a population of both cream-colored colonies and cream-colored colonies with brown, bulls-eyed centers have formed.

Claim 7-8. (Canceled)

Claim 9. (Currently amended) A process of claim [[1]] 35, wherein said chemically-defined medium ~~in step (b)~~ comprises a medium selected from the group consisting of Medium C, Medium D, Medium E, Medium F and Medium G.

Claim 10. (Original) A process of claim 1, wherein said highly productive clonal subtypes of a strain of *E. coli* are cultivated in step (b) on an industrial scale in a chemically-defined medium.

Claim 11. (Original) A process of claim 2, wherein said potential highly productive clonal subtypes are tested in step (b) in a small-scale fermentation system to determine which clonal subtypes are highly productive.

Claim 12. (Original) A process of claim 11, wherein said small-scale fermentation system of step (b) used to test productivity of the potential highly productive clonal subtypes is a shake flask fermentation system using chemically-defined cultivation medium.

Claim 13. (Currently amended) A process of claim 12, wherein said chemically-defined medium comprises KH₂PO₄, K₂HPO₄, (NH₄)₂SO₄ and glycerol ~~DME-B12 medium~~.

Claim 14. (Original) A process of claim 12, wherein a solution is continuously fed to a shake flask containing said potential highly productive clonal subtypes when said clonal subtypes are in mid-logarithmic phase of growth.

Claim 15. (Currently amended) A process of claim 14, wherein said feed solution comprises ~~about 4.6% glycerol (v/v) and about 2.9% monosodium glutamate (w/v)~~.

Claim 16. (Currently amended) A process of claim [[10]] 37, wherein said chemically-defined medium comprises a medium selected from the group consisting of Medium C, Medium D, Medium E, Medium F and Medium G.

Claim 17. (Original) A process of claim 10, wherein said cultivation step comprises at least one production stage fermentation phase.

Claim 18. (Original) A process of claim 17, wherein a solution is continuously fed to a production stage fermentor containing a highly productive clonal subtype when said clonal subtype is in mid-logarithmic phase of growth.

Claim 19. (Currently amended) A process of claim [[18]] 38, wherein said feed solution comprises about 50% glycerol (v/v) and about 25% monosodium glutamate (w/v).

Claim 20. (Currently amended) A process of claim [[18]] 38, wherein said feed solution comprises about 60% glycerol (v/v).

Claim 21. (Original) A method for selecting a highly productive clonal subtype of a strain of *E. coli* for plasmid DNA production comprising the steps of:

(a) purifying colonies of a strain of *E. coli* harboring a DNA plasmid that are phenotypically gray on blood agar, wherein a gray-colored colony represents a potential highly productive clonal subtype; and,

(b) testing productivity of said potential highly productive clonal subtypes, wherein a highly productive clonal subtype exhibits a higher plasmid copy number per cell in comparison to similarly tested *E. coli* clonal subtypes of the same strain.

Claim 22. (Original) A process of claim 21, wherein said strain of *E. coli* is DH5.

Claim 23. (Original) A method of claim 21, wherein said blood agar in step (a) is incubated for about 48 hours at about 30°C.

Claim 24. (Original) A method of claim 21, wherein the productivity of said potential highly productive clonal subtypes of step (b) is determined after cultivating said clonal subtypes in a shake flask fermentation system in chemically-defined medium.

Claim 25. (Currently amended) A method of claim 24, wherein said chemically-defined medium comprises KH₂PO₄, K₂HPO₄, (NH₄)₂SO₄ and glycerol ~~DME-B12 medium~~.

Claim 26. (Original) A method of claim 24, wherein a solution is continuously fed to a shake flask when the potential highly productive clonal subtypes are in mid-logarithmic phase of growth.

Claim 27. (Currently amended) A method of claim 26, wherein said feed solution comprises ~~about 4.6% glycerol (v/v) and about 2.9 % monosodium glutamate (w/v).~~

Claim 28. (Original) A method for selecting a highly productive clonal subtype of a strain of *E. coli* for plasmid DNA production comprising the steps of:

- (a) incubating a strain of *E. coli* harboring a DNA plasmid on chemically-defined agar medium until a population of both cream-colored colonies and cream-colored colonies with brown, bulls-eye centers have formed;
- (b) purifying said cream-colored colonies from step (a), wherein a cream-colored colony represents a potential highly productive clonal subtype;
- (c) testing productivity of said potential highly productive clonal subtypes, wherein a highly productive clonal subtype exhibits a higher plasmid copy number per cell in comparison to similarly tested *E. coli* clonal subtypes of the same strain.

Claim 29. (Original) A process of claim 28, wherein said strain of *E. coli* is DH5.

Claim 30. (Canceled)

Claim 31. (Original) A method of claim 28, wherein the productivity of said potential highly productive clonal subtypes of step (c) is determined after cultivating said clonal subtypes in a shake flask fermentation system in chemically-defined medium.

Claim 32-34. (Canceled)

Claim 35. (New) A method of claim 1, wherein said chemically-defined medium in step (b) comprises KH_2PO_4 , K_2HPO_4 , $(\text{NH}_4)_2\text{SO}_4$ and glycerol.

Claim 36. (New) A method of claim 35, wherein the glycerol concentration in the chemically-defined medium is about 15.0 g/L.

Claim 37. (New) A method of claim 10, wherein said chemically-defined medium comprises KH_2PO_4 , K_2HPO_4 , $(\text{NH}_4)_2\text{SO}_4$ and glycerol.

Claim 38. (New) A process of claim 18, wherein said feed solution comprises glycerol.

Claim 39. (New) A method of claim 24, wherein said chemically-defined medium comprises KH_2PO_4 , K_2HPO_4 , $(\text{NH}_4)_2\text{SO}_4$ and glycerol.